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The Hygienic Quality of Raw milk intended for Human consumption in Algeria: Meta–analysis

La calidad higiénica de la leche cruda destinada al consumo humano en Argelia: Metaanálisis

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ABSTRACT

The main goal of this article was to conduct a meta–analysis, to estimate the hygienic quality of the raw milk intended for human consumption and subsequently relative risk factors that may impact milk quality in Algeria. Weighted random–effects meta–analysis models were employed to analyze the data in this study. A total of 4,685 sampling results from 61 included studies, investigating the bacterial quality of raw milk across 35/48 Provinces in Algeria, from 2009 to 2022. In this meta–analysis, the average level of contamination by Total Aerobic Mesophilic Bacteria (TAMB) was 5.84 $\log_{10}$ CFU·mL^{-1}. 95% CI [5.55–6.13]. The *P*–value was 0.001. The I^2 test indicates the presence of substantial statistical heterogeneity across studies; (I^2=86.26%, *P*<0,001). No significant publication bias was observed using the funnel plots and Egger's test (*P*=0.885). In this article TAMB values did not differ significantly among species (*P*>0.05).The average contamination level TAMB was lower in milk samples collected from individual milking and farm tank milk (4.57 $\log_{10}$ CFU·mL^{-1} and 5.89 $\log_{10}$ CFU·mL^{-1}), compared to those recorded in milk samples collected from the collector, dairy, and sales (6.59 $\log_{10}$ CFU·mL^{-1}, 6.60 $\log_{10}$ CFU·mL^{-1}, and 6.74 $\log_{10}$ CFU·mL^{-1}, respectively). The contamination level of milk by TAMB was higher during summer and spring (6.80 $\log_{10}$ CFU·mL^{-1} and 6.70 $\log_{10}$ CFU·mL^{-1}), compared to winter and autumn (4.27 $\log_{10}$ CFU·mL^{-1} and 5.64 $\log_{10}$ CFU·mL^{-1}).

Key words: Cow's milk; camel's milk; goat's milk; sheep's milk; total aerobic bacteria

RESUMEN

El objetivo principal de este artículo fue llevar a cabo un meta–análisis para estimar la calidad higiénica de la leche cruda destinada al consumo humano y posteriormente identificar los factores de riesgos relativos que pueden afectar la calidad de la leche en Argelia. Se utilizaron modelos de metaanálisis de efectos aleatorios ponderados para analizar los datos en este estudio. Se obtuvieron un total de 4.685 resultados del muestreo de 61 estudios incluidos, que investigaron la calidad bacteriana de la leche cruda en 35/48 provincias de Argelia, desde 2009 hasta 2022. En este metaanálisis, el nivel promedio de contaminación por Bacterias Mesófilas Aerobias Totales (TAMB) fue de 5,84 $\log_{10}$ UFC·mL^{-1}, IC del 95 % [5,55–6,13]. El valor de *P* fue 0,001. La prueba I^2 indica la presencia de una heterogeneidad estadística sustancial entre los estudios (I^2=86,26 %, *P*<0,001). No se observó sesgo significativo de publicación utilizando los gráficos de embudo y la prueba de Egger (*P*=0,885). En este artículo, los valores de TAMB no diferían significativamente entre especies (*P*>0,05). El nivel promedio de contaminación (TAMB) fue más bajo en las muestras de leche recogidas de ordeño individual y de tanques de granja (4,57 $\log_{10}$ UFC·mL^{-1} y 5,89 $\log_{10}$ UFC·mL^{-1}, respectivamente), en comparación con aquellos registrados en muestras de leche recogidas de recolectores, lecherías y ventas (6,59 log10 UFC·mL^{-1}, 6,60 $\log_{10}$ UFC·mL^{-1} y 6,74 $\log_{10}$ UFC·mL^{-1}, respectivamente). El nivel de TAMB fue más alto durante el verano y la primavera (6,80 $\log_{10}$ UFC·mL^{-1} y 6,70 $\log_{10}$ UFC·mL^{-1}), en comparación con el invierno y el otoño (4,27 $\log_{10}$ UFC·mL^{-1} y 5,64 $\log_{10}$ UFC·mL^{-1}).

Palabras clave: Leche de vaca; leche de camello; leche de cabra; leche de oveja; bacterias aeróbicas totales

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INTRODUCTION

Milk has played a major contribution to the human diet in many different Countries across the World since the dawn of time [1]. Nutritional richness is unquestionable; it is a good source of high biological value proteins in addition to important vitamins and essential minerals [2]. Cow's milk is a rich and cheap source of protein and calcium, and a valuable food for bone health [3]. Globally, 16.9% of milk consumed by humans comes from species other than bovine (*Bos taurus*) [4]. There has been an increased interest in Caprine (*Capra hircus*) milk and goat milk products Worldwide because of their high nutritional content and health benefits. Goat milk has various effects on human health considering the total solid, fat, protein, lactose, mineral, and vitamin contents [5]. The beneficial role of Ovine (*Ovis aries*) milk results from its fatty acid, immunoglobulin and non–immune protein contents [6]. Consumption of sheep milk may positively influence the structural integrity of bone, which may result in an enhancement of bone health [7]. The unique physicochemical and biochemical properties of sheep milk also include prebiotics and probiotics which make it perfect functional food for human health promotion and disease risk reduction [6]. Camel (*Camelus bactrianus*) milk showed more nutritional and medicinal properties than other ruminant milks [8], camel milk has anti–microbial and antioxidant properties, camel milk may be a good add–on against breast cancer, liver cancer, human colorectal cancer and hepatitis to relieve the oxidative stress [9].

Composition, nutritional value, and other intrinsic factors, make milk and many milk products attractive for the growth of a variety of microorganisms under suitable conditions [10]. The consumption of raw milk poses a risk to public health due to potential contamination with human pathogenic microorganisms [11]. Moreover bacterial contaminants can cause spoilage of milk and its secondary products [12]. After milking, contaminating microorganisms from equipments and utensils, from environment and even from the employees responsible for obtaining and handling milk, are the most important sources of contamination [13].

With an average consumption of 130 L of milk per person per year, Algeria is the largest consumer of milk in the Maghreb Region [14]. Local milk production estimated at 2.5 billion L was provided largely (nearly 80%) by cattle [15], the rest is represented by sheep, goat, however camel milk production is very marginal (FIG. 1) [16]. Over the past decades, several studies have been published on the hygienic quality of milk produced in Algeria, but the results are highly divergent and have been obtained under highly variable conditions.

The objective pursued in this article, therefore, to perform a synthesis of these data through a meta–analysis. And subsequently to estimate the relative risk factors that can affect milk quality in Algeria.

Meta–analysis can be a valuable tool for summarizing research findings across studies. It permits reviewers to describe the results of each study on a common effect size metric, combine information from many studies in an optimal fashion, and understand the degree to which the findings from different studies agree with one another [17].

MATERIALS AND METHODS

To conduct this meta–analysis, the guidelines provided by PRISMA were adhered (Preferred Reporting Items for Systematic Reviews and Meta–analyses).

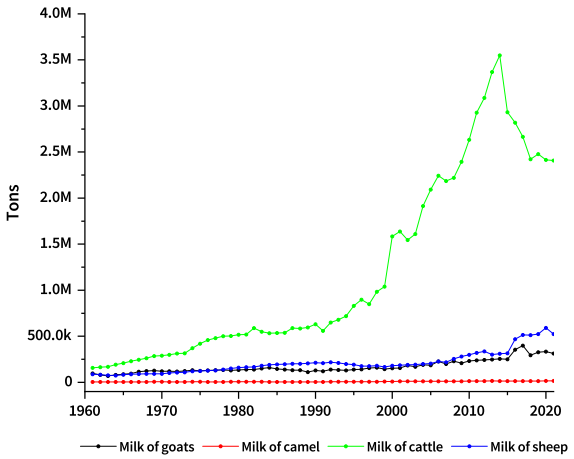

FIGURE 1. Annual milk production from different lactating mammals intended for human consumption in Algeria (1961–2021). Source FAOSTAT (Mar 30,2023)

Data collection

The aim of this systematic literature review was to conduct a meta–analysis, to investigate the potential risk factors that may influence milk quality in Algeria.

It was conducted a search for studies published in specific repositories and indexed sources (Google Scholar, Pubmed, Science Direct), as well as both undergraduate and graduate degree works in Algerian University repositories. The search query employed in two languages French and English, was as follows: "Milk and cattle, goat, sheep, camel and hygiene, TAMB (Total Aerobic Mesophilic Bacteria) and Quality and Algeria". A thorough and comprehensive search across all available data bases was performed.

Multiple criteria were employed to determine the inclusion of eligible studies: it was included studies that allowed for the detection of TAMB in raw milk, focusing on its hygienic characteristics collected from different stage of production (Farm, Dairy, Sale), and during different seasons (Spring, Summer, Autumn, Winter). The search executed on 09/12/2022 with the final update performed on 28/02/2023, yielded 332 studies. Duplicate entries were subsequently removed, and an initial screening was conducted based on a preliminary review of the study titles. Pasteurized milk, Powder milk, Cheese, Yogurt and Whey studies were excluded from consideration. Following this initial screening, a total of 96 studies were selected for further analysis. A second screening was conducted to identify 61 eligible articles that were ultimately included in the meta–analysis. Publications that lacked clarity in their methodology or results; those without any statistical or quantitative contributions and those with incomplete data were excluded from consideration.

A flowchart was created to summarize the step–by–step process of study selection in the meta–analysis. This flowchart provides a visual representation of how studies were identified, screened, and included or excluded at each stage of the selection process (FIG. 2).

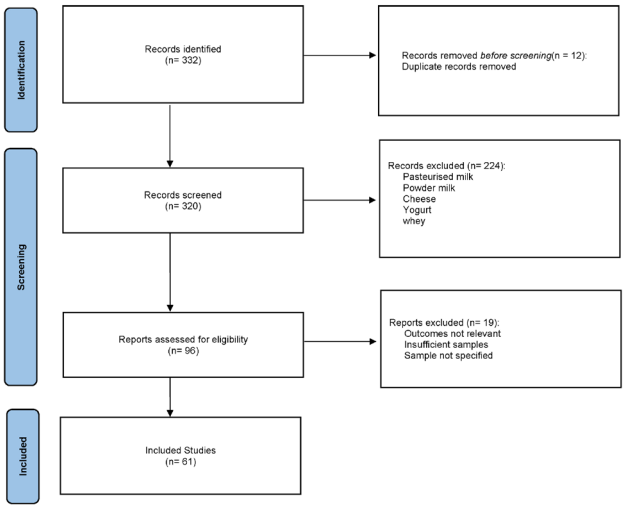

FIGURE 2. Flow chart summarizing the process of study selection

Statistical analysis

Meta–analysis can be a valuable tool for summarizing research findings across studies [17]. Outcomes from a meta–analysis may include a more precise estimate of the risk factor, or other outcomes, than any individual study contributing to the pooled analysis [18]. Weighted random–effects meta–analysis models were employed to analyze the data in this study. The results of meta–analyses are often presented in a forest plot. Heterogeneity of results among trials is quantified using the inconsistency index I^2.Funnel plots and the Egger's test were used to assess publication bias. The statistical significance level was set at $P<0.05$, R (version 3.5.1) software was used for all analyses.

RESULTS AND DISCUSSION

Milk has a composition that creates favorable conditions for the growth of various microorganisms. However, this can pose a risk to public health. Additionally, bacterial contaminants can lead to spoilage of milk and its derived products.

This meta–analysis was conducted to examine milk quality produced from different geographical areas in Algeria, intended for human consumption. Milk contamination level was determined by the TAMB method at 30°C, for each sample of milk. Meta–analysis is a statistical method to combine results of different studies, especially those with small sample size or with conflicting results [19].

The mesophilic microorganism is ones of the more general and extensively microbiological indicators of food quality, indicating the adequacy of temperature and sanitation control during processing, transport, and storage, and revealing sources of contamination during manufacture [20]. The total aerobic count is the most common method for evaluation of bacterial quality of raw milk. This count estimates the total number of bacteria present in raw milk at the time of pickup. It provides an overall measure of hygienic quality of milk [21]. The legal total bacterial counts limit in farm raw milk is set at 100,000 cells·mL^{-1}., across the Europe and the Americas [12]. Local limits on TAMB in raw milk for human consumption range from less than or equal to 3.10^5 CFU·mL^{-1} to less than or equal to 3.10^6 CFU·mL^{-1} [22]. When total bacterial counts in grade A unpasteurized milk is less than the regulatory limit of 100,000 CFU·mL^{-1}, it is assumed that all pathogenic and most nonpathogenic bacteria are destroyed by pasteurization [23].

Study identification

It was collected a total of 4,685 sampling results from 61 studies investigating the bacterial quality (TAMB) of raw milk across 35/48 Provinces in Algeria from 2009 to 2022 (FIG. 3).

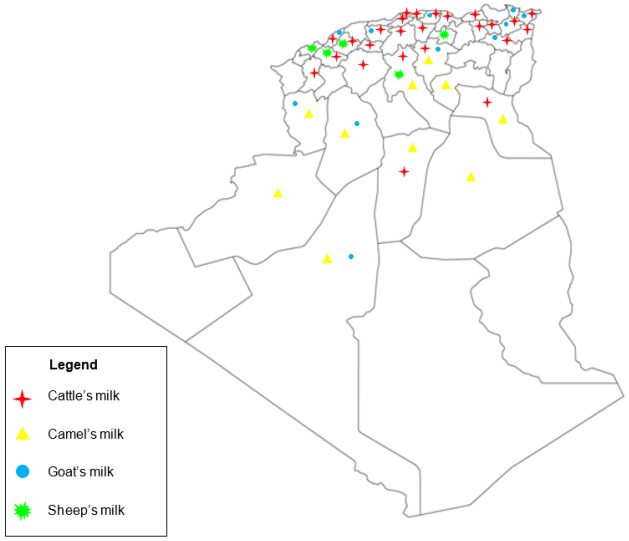

FIGURE 3. Map of locations of studies included in the meta–analysis. Algerian map was downloaded from Vemaps.com

Publication bias

No significant publication bias was observed using the funnel plots for asymmetry (FIG.4) and Egger's test (P= 0.885).

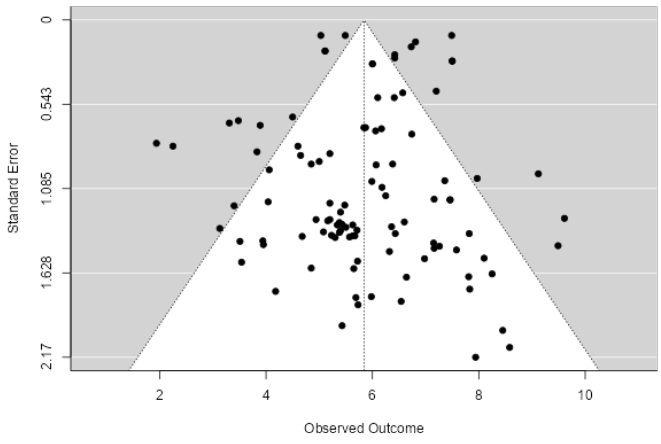

FIGURE 4. Funnel plot for the Total Aerobic Mesophilic Bacteria (TAMB) level

Assessment of heterogeneity

The I^2 test indicates the presence of substantial statistical heterogeneity across studies; (I^2 = 86.26 %, degrees of freedom [d.f.] = 102, P< 0.001). The conduct of a meta–analysis is therefore justified. Thus, in an effort to reduce this heterogeneity, it was performed subgroup analyses to explore factors that may explain the heterogeneity.

Overall results of the studies

In this meta–analysis, the average level of contamination by TAMB was 5.84 log_{10} CFU·mL^{-1}. 95% CI [5.55–6.13]. The P–value was 0.001 for a total of 4,685 samples from 61 included studies are presented in [24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84] (FIG. 5).

The summarized results of the meta–analysis about the level of contamination by TAMB found in included studies, are presented in tabular form (TABLEI), concerning different species (Cow, Camel, Goat, Sheep), stage of production (Farm, Collector, Dairy unit, Local market), seasons (Winter, Spring, Summer, Autumn). It also displays the number of studies analyzed, the total number of samples, and the heterogeneity (I^2). Moreover, four forest plots are provided as examples (FIGS.5, 6, 7 and 8).

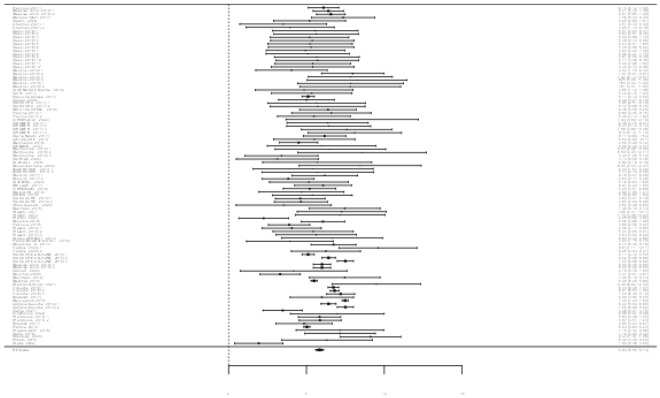

FIGURE 5. Forest plot of the overall studies

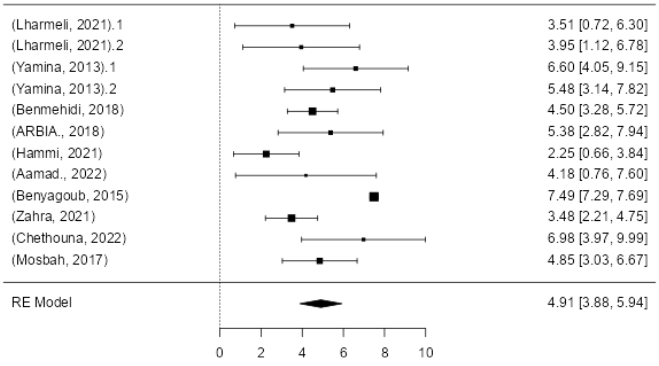

FIGURE 6. Forest plot of the bacterial load of the Camel's raw milk

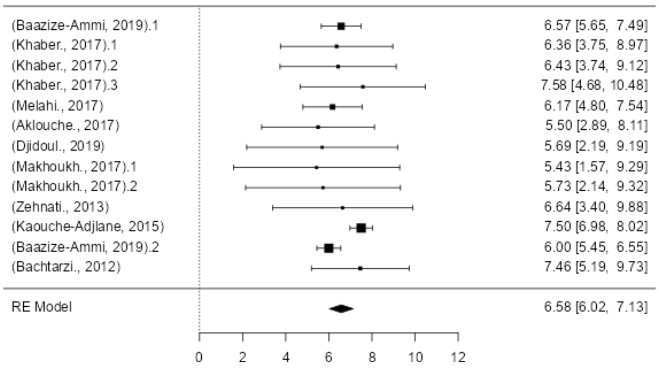

FIGURE 7. Forest plot of the bacterial load of the Dairy's milk

TABLE I
Summarized results of the meta–analysis

Variable	Level	Number of Observations / Sample	I^2 (%)	TAMB (log_{10} CFU·mL^{-1})	IC 95%	P–value
Species	Cow's milk	71/4223	72.79	6.05	5.78–6.32	0.620
	Camel' milk	12/166	80.47	4.91	3.88–5.93	
	Goat's milk	17/135	72.56	5.82	4.67–6.93	
	Sheep's milk	3/161	60.85	5.60	4.09–7.09	
Production step	Individual	26/415	65.62	4.57	3.99–5.14	0.001
	Farm (tank)	46/2931	69.36	5.89	5.52–6.26	
	Collector	3/155	45.09	6.59	6.21–6.95	
	Dairy Unit	13/512	38.93	6.60	6.02–7.13	
	Local Market	33/555	70.48	6.74	6.16–7.32	
Season	Winter	4/105	28.14	4.27	2.99–5.58	0.045
	Spring	12/333	29.86	6.70	6.04–7.36	
	Summer	3/201	0.01	6.80	6.59–7.01	
	Autumn	4/47	0.01	5.64	4.32–6.95	

Source: Research data

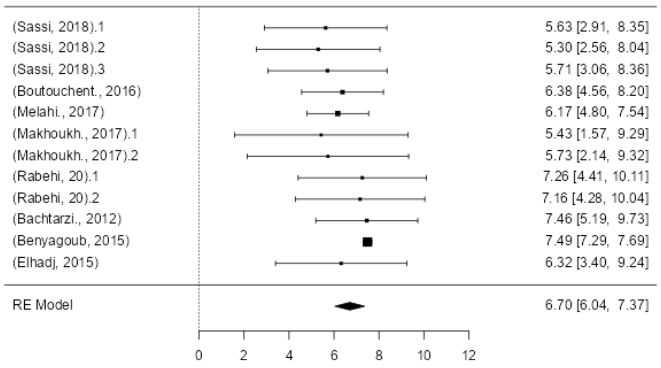

FIGURE 8. Forest plot of the contamination level in raw milk by Total aerobic mesophilic bacteria (TAMB) during spring

The hygienic quality of milk according to the species

Studies focusing on sheep showed low heterogeneity (I^2 = 60.85%), whereas studies on Camel, Cow, and Goat exhibited higher heterogeneity (I^2 = 80.47, 72.79 and 72.56%, respectively). The average contamination level measured by TAMB was lower (4.91 $\log_{10}$ CFU·mL^{-1}) in camel milk (FIG. 6), compared to the average microbial contamination level in sheep, goat, and bovine milk (5.60 $\log_{10}$ CFU·mL^{-1}, 5.82 $\log_{10}$ CFU·mL^{-1}, 6.05 $\log_{10}$ CFU·mL^{-1}, respectively).

In this article, TAMB values did not differ significantly (P-value>0.05) among species. Similar results were conducted by Verraes *et al*., [11], who conclude that there is no significant difference in the total plate count results of raw milk from goats, sheep and cows. It should be noted that these frequencies can be explained by the season in which the samples were taken, the size of the farm, the density of the animal population, regional differences in the keeping and taking care of animals [11].

Total bacteria count in raw milk at different stages in the dairy chain

Heterogeneity was very high in studies addressing the hygienic quality of milk samples collected at the Local Market level (I^2 = 70.84%), whereas studies focusing on milk samples from other production stages (Dairy Unit, Collector, Individual Milking, Farm's milk from tank) showed lower heterogeneity rates (I^2= 38.93, 45.09, 65.62 and 69.36% respectively). The TAMB was lower (4.57 $\log_{10}$ CFU·mL^{-1} and 5.89 $\log_{10}$ CFU·mL^{-1}) in milk samples collected from individual milking and farm tank milk (FIG. 7), compared to those recorded in milk samples collected from the collector, dairy, and sales (6.59 $\log_{10}$ CFU·mL^{-1}, 6.74 $\log_{10}$ CFU·mL^{-1}, and 6.75 $\log_{10}$ CFU·mL^{-1}, respectively).

Raw milk quality at farm level is an important component influencing the performance of the whole dairy chain [85]. It was concluded that milk from the cow is of good hygienic quality, but milk is often contaminated after milking, and the hygienic quality is very low when it reaches the consumers [86]. The deterioration in milk hygiene quality between cow and farm tank level was probably due to contaminated milking vessels and tank milk containers [86]. Improper and inadequate cleaning and sanitizing of milking equipment (e.g., milking machine, bulk tank) is probably the most significant source of environmental contamination of milk. In addition, equipment of poor sanitary design, or that has not been maintained, contribute to microbial contamination of the milk.

Other factors related to refrigerated storage (storage temperature, and storage time) are important to minimizing their growth [10]. The high total bacteria count in milk from milk collection center suggests proliferation or recontamination of milk by bacteria during transportation [87]. The hygienic quality of the milk, shown as total bacteria count, became so much worse during storage already at the farm and continued to decrease a further along the dairy chain [86].

Bacterial quality of milk according to the season

Heterogeneity was absent in studies focusing on milk samples collected during Summer, and Autumn. Studies involving milk samples collected during Spring and Winter showed higher heterogeneity (I^2 = 29.86%, I^2 = 28.14). The contamination level of milk by TAMB was higher during Spring (FIG. 8) and Summer (6.70 $\log_{10}$ CFU·mL^{-1} and 6.80 $\log_{10}$ CFU·mL^{-1}, respectively), compared to Winter and Autumn (4.27 $\log_{10}$ CFU·mL^{-1} and 5.64 $\log_{10}$ CFU·mL^{-1}).

The season also plays a significant role; fluctuations in temperature can exert significant influence on bacterial counts in raw milk. TAMB values were significantly affected by sampling month. Seasonal variation of TBC, with high values during Summer and Spring, and lower values during Winter, is consistent with previous studies. Similar results were reported by Zucali *et al*. [85] in a study in dairy farms situated in the North of Italy (Lombardy), who observed strong seasonal variations in total aerobic counts, with higher total aerobic counts in the Summer. Season affects total aerobic count with a positive trend during Summer and an opposite one in Winter, as shown by Elmoslemany *et al*. [21] in a study on dairy herds of Prince Edward Island, who concluded that higher milk bacterial counts during Summer and Spring, may be related to warmer ambient temperature, allowing bacteria to grow faster.

Good hygiene practices during milking and subsequent handling of milk are essential to reduce the risk of contamination on the farm and in the milk processing plant [88]. Two alternative approaches are observed Worldwide to ensure the sanitary safety of milk: in the United States of America, emphasis is placed on control and sterilization, while in Europe; the focus is on quality and safety management throughout the supply chain. The latter approach seems more appropriate for Southern Countries where state control systems are weak and the risks of contamination are numerous [4].

CONCLUSIONS

In conclusion, the meta-analysis conducted in this study sheds light on the bacterial quality of raw milk intended for human consumption in Algeria. The findings reveal an average contamination level of total aerobic bacteria (TAMB) at 5.84 $\log_{10}$ CFU·mL^{-1}, with a statistically significant presence of heterogeneity among the included studies. Despite this variation, no significant publication bias was observed.

The analysis also underscores the impact of different factors on milk quality. The source of milk collection exhibited noteworthy differences, with lower TAMB levels observed in samples collected from individual milking and farm tanks compared to those obtained from collectors, dairies, and sales. Additionally, seasonal variations played a role, as higher contamination levels were recorded during the summer and spring months compared to the winter and autumn seasons.

It is noteworthy that TAMB values did not show significant variations among different species, highlighting the importance of monitoring and improving overall hygiene practices throughout

the milk production process. These results emphasize the need for consistent efforts to enhance the hygienic quality of raw milk in Algeria and suggest potential areas for targeted interventions, such as during specific seasons and stages of milk collection.

As future research progresses, these insights can contribute to the development of effective strategies to ensure the safety and quality of raw milk for human consumption. It is evident that ongoing monitoring, research, and collaboration among stakeholders are crucial to maintaining and improving the bacterial quality of raw milk, thereby safeguarding public health and promoting the well-being of consumers.

Conflicts of interest

The authors declare no conflict of interest.

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